

U.S.S.N. 09/909,574

Filed: July 20, 2001

AMENDMENT AND RESPONSE TO OFFICE ACTION

In the Claims

Please enter the following amendment.

1. (once amended) A method for producing polyhydroxyalkanoates comprising providing ~~genetically engineered~~ organisms which express enzymes selected from the group consisting of ~~diol oxidoreductase, aldehyde dehydrogenase,~~ acyl-CoA transferase, acyl-CoA synthetase, β -ketothiolase, acetoacetyl-CoA reductase, and PHA synthase, providing diols which can be converted into hydroxyalkanoate monomers by enzymes expressed ~~by~~ in the organisms, wherein
- the organisms are transformed with genes encoding enzymes selected from the group consisting of diol oxidoreductase and aldehyde dehydrogenase, wherein
- diol oxidoreductase and aldehyde dehydrogenase convert 1,4 butanediol into hydroxyalkanoate monomers, and
- culturing the organisms under conditions wherein the hydroxyalkanoate monomers are polymerized to form polyhydroxyalkanoates.
2. (original) The method of claim 1 wherein the diol is 1,6-hexanediol and the hydroxyalkanoate monomer is 6-hydroxyhexanoate.
3. (original) The method of claim 1 wherein the diol is 1,5-pentanediol and the hydroxyalkanoate monomer is 5-hydroxyvalerate.
4. (original) The method of claim 1 wherein the diol is 1,4-butanediol and the hydroxyalkanoate is 4-hydroxybutyrate.

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5. (original) The method of claim 1 wherein the diol is 1,3-propanediol and the hydroxyalkanoate monomer is 3-hydroxypropionate.

6. (original) The method of claim 1 wherein the diol is 1,2-ethanediol and the hydroxyalkanoate is 2-hydroxyethanoate.

7. (original) The method of claim 1 wherein the diol is 1,2-propanediol and the hydroxyalkanoate is 2-hydroxypropionate.

8. (original) A genetically engineered organism for use in the method of claim 1 comprising an organism which expresses the *aldH* and *dhaT* genes.

9. (original) The organism of claim 8 wherein the organism is selected from the group consisting of *Escherichia coli*, *Ralstonia eutropha*, *Klebsiella* spp., *Alcaligenes latus*, *Azotobacter* spp., and *Comamonas* spp.

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10. (once amended) A system for making polyhydroxyalkanoates comprising an organism genetically engineered to express enzymes selected from the group consisting of a diol oxidoreductase, ~~aldehyde dehydrogenase~~, acyl-CoA transferase, acyl-CoA synthetase, β -ketothiolase, acetoacetyl-CoA reductase, and PHA synthase, wherein the organism is transformed with genes encoding enzymes selected from the group consisting of diol oxidoreductase and aldehyde dehydrogenase, and wherein the organism can convert diols into hydroxyalkanoate monomers which are polymerized to form polyhydroxyalkanoates

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~~wherein the organism can convert diols into hydroxyalkanoate monomers which are polymerized to form polyhydroxyalkanoates.~~

11. (original) A composition comprising a polyhydroxyalkanoate copolymer which includes

2-hydroxypropionate or 2-hydroxyethanoate or both, and

at least one comonomer selected from the group consisting of 3-hydroxybutyrate, 4-

hydroxybutyrate, 3-hydroxypropionate, 2-hydroxybutyrate, 4-hydroxyvalerate, 5-

hydroxyvalerate, 6-hydroxyhexanoate, and 3-hydroxyhexanoate, having a weight-average

molecular weight (Mw) of at least 300,000.

12. (Once amended) The composition of example claim 11 where the comonomer is 3-hydroxybutyrate.

13. (Once amended) The composition of example claim 11 where the comonomer is 4-hydroxybutyrate.

14. (Once amended) The composition of example claim 11 where the comonomer is 3-hydroxypropionate.

15. (Once amended) The composition of example claim 11 where the comonomer is 2-hydroxybutyrate.

16. (Once amended) The composition of example claim 11 where the comonomer is 4-hydroxyvalerate.

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17. (Once amended) The composition of ~~example~~ claim 11 where the comonomer is 5-hydroxyvalerate.
18. (Once amended) The composition of ~~example~~ claim 11 where the comonomer is 6-hydroxyhexanoate.
19. (Once amended) The composition of ~~example~~ claim 11 where the comonomer is 3-hydroxyhexanoate.
20. (original) A method for improving a biological system for making polyhydroxyalkanoates with an organism genetically engineered to express enzymes selected from the group consisting of a diol oxidoreductase, aldehyde dehydrogenase, acyl-CoA transferase, acyl-CoA synthetase, β -ketothiolase, acetoacetyl-CoA reductase, and PHA synthase, wherein the organism can convert diols into hydroxyalkanoate monomers which are polymerized to form polyhydroxyalkanoates, the method comprising selecting for mutants with increased enzyme activities by
- i) introducing mutations into a specific host, and
 - ii) screening pools of the mutants generated for increased ability to synthesize PHA from a selected diol or diols.
21. (original) A DNA fragment encoding a diol oxidoreductase and an aldehyde dehydrogenase, wherein the expressed enzymes can produce hydroxyalkanoate monomer selected from the group consisting of 3-hydroxybutyrate, 4-hydroxybutyrate, 3-hydroxypropionate, 2-hydroxybutyrate, 4-hydroxyvalerate, 5-hydroxyvalerate, 6-

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a' hydroxyhexanoate, 3-hydroxyhexanoate, 2-hydroxypropionate, and 2-hydroxyethanoate from diol.
